EXPLORATION AND CATALOGING OF DIFFERENT PLANTS FROM HIMALAYAN REGION FOR THEIR THERAPEUTIC ACTIVITY

Dissertation Submitted in partial fulfillment of the requirement for the degree of

Bachelor of Technology

In

Biotechnology

By - Naveeta Karar

Roll no- 171820

Under the Supervision of

Dr. Jitendraa Vashistt

To

JAYPEE UNIVERSITY OF INFORMATION TECHNOLOGY, WAKNAGHAT

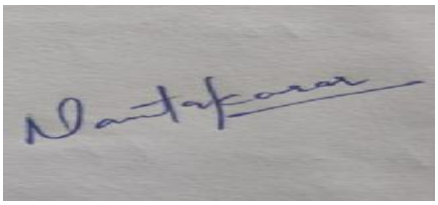
DEPT. OF BIOTECHNOLOGY AND BIOINFORMATICS

HP-173234, INDIA

DECLARATION

I hereby declare that the work reported in the B. Tech. thesis entitled "**Exploration and Cataloging of different plants from Himalayan region for their therapeutic activity** " submitted to Jaypee University of Information Technology, Waknaghat, India, is an authentic record of our work carried out under the supervision of **Dr. Jitendraa Vashistt**, Associate Professor, Dept. of Biotechnology and Bioinformatics, JUIT, Waknaghat, HP-173234, India during July 2020 to May 2021.

I also declare that no part of this thesis has previously been submitted to any University or any examining body for acquiring any degree.



(Naveeta karar, 171820)

Department of Biotechnology and Bioinformatics,

Jaypee University of Information Technology, Waknaghat, Solan, Himachal Pradesh, India.

Date: 22/05/2021

CERTIFICATE

This is to certify that the work reported in the B. Tech. thesis entitled "**Exploration and Cataloging of different plants from Himalayan region for their therapeutic activity**" ,submitted by Naveeta karar (171820) at Jaypee University of Information Technology, Waknaghat, India, is a bonafide record of his original work carried out under my supervision. This work has not been submitted elsewhere for any other degree or diploma.

(Dr. Jitendraa Vashistt)

Dept. of Biotechnology and Bioinformatics,

Jaypee University of Information Technology (JUIT), Waknaghat, Solan.

Himachal Pradesh, India – 173234

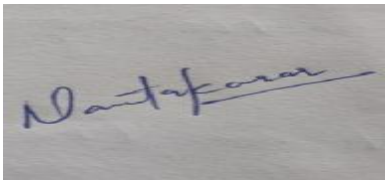
Date: 22/05/2021

ACKNOWLEDGEMENT

Department of Biotechnology and Bioinformatics at Jaypee University of Information Technology, Waknaghat for providing all the students of biotechnology an opportunity to choose desirable individual study and assigning well knowledge faculty to guide the students throughout their project work.

I extend my deep sense of gratitude towards my project guide Dr. Jitendraa Vashistt, Associate Professor, Department of Biotechnology and Bioinformatics at Jaypee University of Information Technology for his invaluable guidance, keen interest and inspiration which helped me in carrying out the project work. He steered me in the right direction whenever I came across any difficulty throughout the work.

I am grateful to my parents for providing continuous encouragement throughout my period of study.



Naveeta Karar (171820)

TABLE OF CONTENTS

CHAPTER 5- CONCLUSION

CHAPTER 6- REFERENCES

ABSTRACT

The inhabitants of the hilly areas of the Indian Himalaya depend heavily on plants to treat various diseases. The indegenous knowledge and traditional practices of medicinal plants are vanishing fast. Because of better compliance and better cultural acceptibility with the human body and fewer side effects, herbal medicine is still the mainstay of about 75 to 85 percent of the world's population, mostly in developing countries, for primary health care. Local healers are knowledgeable about a broad range of medicinal plants that can be used to treat of ailments. They emphasise cures for stomach problems, skin diseases, fevers, and respiratory infections, among other things. The Indian Himalayan Region is one of the richest reservoirs of biological diversity in the world and it is considered as the store house of the valuable medicinal plant species. The medicinal plants were local, and the reported species included trees, herbs, shrubs, and fungi. Various plant parts were used to treat various diseases, including leaves, roots, fruits, tubers, seeds, fruiting body, stem, wood, flowers, and bark. Bioactive compounds such as amides, flavonoids, saponins, alkaloids, terpenoids, glycosides, are widely utilized to be present in the leaves, seeds and stem bark. The application of traditional phytochemical screening assays, chromatographic techniques such as TLC and HPLC and non-chromatographic techniques, to the study of bioactive compounds present in plant extracts. As a result, a growing number of researchers are turning their attention to traditional medicines and attempting to create better antimicrobial drugs. Several medicinal plant extracts are highly effective against microbial and parasitic infections.

Keywords: medicinal plants, bioactive compounds, antimicrobial activities.

Chapter 1

INTRODUCTION

In the Indian Himalaya, Himachal Pradesh has a diverse range of medicinal plant that are commonly used. It spans over 2,800 kilometres and is 220 to 300 kilometres high, with altitudes ranging from 200 to 8000 metres. There are 1748 medicinal plant species with numerous modern therapeutic and traditional applications, as well as 118 medicinal plant species that produce essential oils. A large number of medicinal plants have been carried out in the IHR. The IHR has conducted research on a large number of medicinal plants [1]. Ayurvedic, Unani, and other conventional medical systems, as well as plant-based pharmaceutical industries, use medicinal plants. According to estimates, 90% of medicinal plant species are harvested from the wild, with 69 percent collected by destructive harvesting [2]. Himachal Pradesh has a diverse biodiversity that is both natural and representative, as well as socioeconomically significant. It spans a wide altitude range (200-7109m), with a diverse range of animals, populations, and ecosystems. Tropical vegetation, which includes both deciduous and evergreen forests, is found in the lower parts of the states. Evergreen forests dominate subtropical vegetation, which ranges in altitude from 500 to 1800 metres. Subalpine vegetation ranges from 2802 to 3800 metres, and alpine vegetation ranges from 3800 metres. Plants have been used by local people for a variety of purposes, including medicine [3]. They generate income by trading some of the most valuable medicinal plants.The temperate region, which is less than 1800 metres above sea level, has the greatest number of medicinal plants (1801-2800m). Conservation of habitats, plants, and ecosystems has been a priority for both state and federal governments [4]. Altitude is the most significant factors influencing the distribution of a medicinal plants in the state high-volume and low value species are found at low altitudes, whereas high-value species extract in limited amounts are found at high altitudes. Majority of medicinal herbs are concentrated at high altitudes, while small forest products are found at lower elevations [5]. The most important requirement in medicinal plants is long-term extraction. Agriculture and ayurveda in Himachal Pradesh are focusing on processing and extraction techniques. Purification of

active compounds from the medicinal plants and other natural sources, as well as synthetic chemistry, have all been used to obtain compounds for drug development. Mostly the plant medicines have been used in their crude forms [6].

Herbal medicines effectiveness: Herbal medicines are thought to be safe and, in most cases, unique in their function to organs and systems of the human body, with the assumption that they can be used to treat diseases where traditional medicine fails. Synthetic and chemical medications have more potent and faster effects than herbal medicines, but they come with a slew of risks and side effects. Synthetic drugs are highly cost: Medicinal plants continue to make a significant contribution to current prescription medicines by supplying constituents that can be used to create new drugs. In recent years, the quest for and use of medicines and dietary supplements has been accelerated using medicinal plants [7].

Chapter 2

REVIEW OF LITERATURE

A medicinal plant is any of a number of plants that are used in herbal medicine. It encompasses both the practise of utilising plants for therapeutic purposes and the research of such practises. The word herb comes from the Latin word herba. Herbs can now refer to any component of a plant, including the stem, seed, fruit, leaves, and bark, as well as non-woody plants like those found on trees and shrubs. These medicinal plants are also used as food, a source of flavonoids, medicine, and for spiritual purposes. Traditional medical services continue to be widely used. Increased focus on the use of plant products as a treatment for infectious diseases has resulted from an insufficient supply of medications, prohibitive treatment costs, side effects, and the emergence of resistance to currently used drugs for infectious diseases have led to increased emphasis on the use of plant materials as a source of medicines for a wide variety of human ailments [8].

There are over 21,000 plant species that have therapeutic potential. Plants and plant extracts are the primary source of health treatment for almost three-quarters of the world's population, according to current research. More than 30% of all plant species have been utilised medicinally at some point in their history. Plant medicines make up to 25% of all pharmaceuticals. Economic value of the medicinal plants is even greater in India than the rest of the world. Use of medicinal plants is thought to be very healthy, as there are few side effects. The fact that these remedies are in tune with nature is the greatest benefit. Aloe, Tulsi, Neem, Turmeric, and Ginger are medicinal plants that can help with a variety of ailments. In several parts of the world, these are considered home remedies.

Herbs are utilised in natural colouring, fruit, pest control, and tea. Pharmaceutical manufacturing currently relies heavily on medicinal plants. Traditional medicine practitioners provide highly efficient recipes for the treatment of common diseases such as diarrhoea, constipation, hypertension, bronchial asthma, and fever [9].

Importance of Medicinal plants:

Several plants have medicinal properties which can be used as medicines:

Rauwolfia- This plant used for the treatment of insomnia and hypertension.

Belladona- Alkaloids extracted from the roots of the plant are used to treat pain and promote respiration and circulation.

Eucalyptus- The leaves of the eucalyptus tree are used to extract oil, which is used to treat blocked nose and throat infections and as a mosquito repellent.

Disinfectant properties are found in several therapeutic plants, which kill disease-causing bacteria. They also stop harmful bacteria from multiplying and causing illness.

Calming herbs, which have a calming impact on the body, were suggested by medicinal plants. Sedatives are commonly utilised [10].

Antifungal agents: are used to treat infections caused by fungi. Also known as mycoses.

Broken down into molds and yeast.

Yeast- Reproduce by budding and Single-cell fungi.

Molds- Multicellular and branching filaments called hyphae.

Various mechanism for antifungal agents:

1) Polyenes- nystatin and amphotericin B.[11]

   Bind to sterols in the cell membrane lining, allowing k+ and Mg++ to leak out, changing the metabolism of fungal cells.

   Result- fungal cell death.

2) Flucytosine

   Flucytosine (antimetabolite), also known as 5-fluorocytosine, is taken in by fungal cells and interferes with DNA synthesis.

   Result- fungal cell death.

3) Imidazoles

   Imidazoles block an enzyme, causing cell membrane leakage and a change in cell membrane.

   Result- fungal cell death.

ASSAY AND DETECTION

2.1 Antimicrobial assay

Antimicrobial agents are the compounds that have the ability of either killing the microorganisms or of stopping their growth. An antimicrobial can be an antifungal (against fungi), antibacterial/antibiotic (against bacteria), antiparasitic (against parasites). Agents that kill the microbes are called microbicidal and those that only inhibit the growth are known as biostatic. [12]

2.1.1 Antimicrobial Susceptibility test

Also abbreviated as AST, it is a test used to determine the specificity of antibiotic towards a group of organism. It is a procedure in the clinical microbiology laboratory. It determines at what concentration does the antimicrobial inhibits the microbial growth in vitro. The most common method of AST antimicrobial susceptibility test is the disk diffusion method [12], [13].

2.1.2 Antimicrobial gradient method

Antimicrobial concentration gradient in the agar medium as the means of determining the susceptibility. [14]

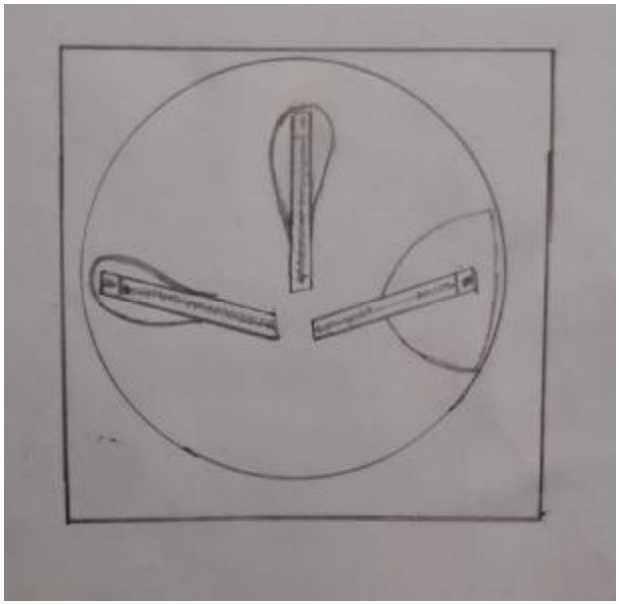


Figure2: Determines the minimum inhibitory concentration of each agent and intersection of the organism growth with the strip as measured and using the scale inscribed on the strip.

2.1.3 Disk diffusion test

Disk diffusion method is performed by applying the bacteria inoculums that is to be Mueller-Hinton agar plate tested. On the agar plate are placed the paper and antibiotic discs. After incubation the zones of growth inhibition are around each disk is measure very precisely. The zone's diameter is proportional to the isolate's susceptibility and the drug's rate of diffusion through the agar media. The result are reported as MIC (minimal inhibitory concentration) that is low concentration of the compound, and that inhibit the growth of the microorganism in the test. It provides a quantitative assessment of the dosage. [15]

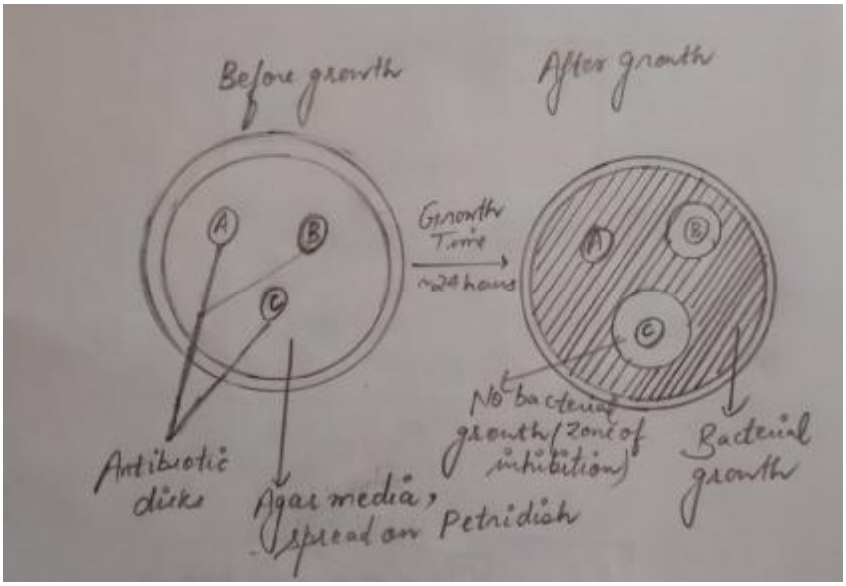


Figure3: Disk diffusion test before and after the diffusion.

2.1.4 Poisoned food method

The most common use of the poisoned food method is to assess the antifungal effect against moulds. The antifungal agent or extract is mixed thoroughly into molten agar at the desired final concentration. The medium is then poured into Petri dishes to continue the experiment. Inoculation can be achieved with a mycelia disc ranging from 2 to 5 mm, which is deposited in the centre of the plate after an overnight pre-incubation. Diameter and sample plates are determined after further incubation under conditions appropriate for the fungal strain studied, and the antifungal effect is calculated by:

Antifungal activity(%) =((Dc-Ds)/Dc)*100

Where Dc is the diameter of growth in control plate and Ds is the diameter of growth in the plate containing tested antifungal agent. Sporulation can be also compared to the control. [16]

2.1.5 Time kill test

Time kill test, also known as time kill curve, is used to determine the test compound's fungicidal and bactericidal impact. The test can be performed in two ways: time-dependent or concentration-dependent. CFU/ ml stands for colony forming unit per ml of medium, and it is used to calculate the percentage of dead cells at various concentrations and time intervals.

Chapter 3

MATERIALS AND METHODS

Objective 3.1 : To screen and categorized different medicinal plants based on the altitude they are found in, bioactive compounds and their therapeutic properties.

Procedure: Different Plants were screened for their different medicinal properties. Antifungal activity and bioactive compounds responsible for the same was also noted [17],[18],[19],[20].

Table1:

Botanical name of the plant	Antifungal activity	Altitude and temperature	Alkaloid compound	Flavonoids compound	Another chemical compound
Allium sativum	+	Temp between 0 to 10 deg C	_	+	Vinyldithiins
Althaea cannabina	_	Altitude of 0-800m	_	+	Phenolic acids, sucrose, asparagine.
Allamanda schotii	_		_	+	_
Aloe barbadensis	+	Annual temp within the range19-27 deg C, temp range 10-35 deg C	_	+	Caffeic acids
Aconitum ferox	+	1.0m tall by 0.5m wide and tolerant	+	_	Indaconitine, Hypoaconitine
Alstonia scholaris	_	Temp 12-32 deg C	_	_	Stigmasterol, beta-sterol
Bacopa monnieri	+	Altitude upto 1300m, temp 15-40 deg C	+	_	Nicotinine, betulinic acid
Bauhinia	+		_	+	Tannins

vahlii					
Curcuma longa	+		_	_	Polyphenolic
Cinnamous Zeylanicum	+	Low altitude, temperature of about 27 deg C	_	_	Cinnamyl acetate
Coriandrum sativum	+	1.3m tall upto 2cm in diameter, temp above 4 deg C	_	_	Dodecanal, alpha-pinene
Citrus limon L.	+	Altitude upto 1200m, temp range of 13-37 deg C	_	+	Sesquiterpenes, citronellal
Carica papaya	+	Temp range 21-33 deg C being 25 deg C with the optimum value	+	_	Pseudocarpaine
Camellia sinensis	+	Temp range between 18-20 deg C	_	_	Glycosides, terpenoids
Dillenia indica L.	+	Annual daytime temp 30-40 deg C	+	+	Saponins, tannins
Dolichos biflorus	+		+	+	Tannins
Datura metel	+	Temp 10 deg C	_	_	Scopolamine, adenosine
Eucaplytus globules	_	Temperature maximum of 45 deg C	_	_	Alpha-pinene,limonene
Eclipta alba (L). Hassk	+	Annual herb upto 30-40cm high	_	+	Terpenoids

Ficus carica	+	Large shrubs that grows upto 7- 10m	+	_	Terpenoids, eugenol
Ficus racemosa	_	20-30m tall	_	_	Tannins, saponins
Gmelina arborea	+	Annual temp range 21-28 deg C	_	+	lignans, flavones glycoside
Hibiscus rosa-sinensis L.	+	2.5-5m tall, not tolerate temp between 10 deg C	+	+	Steroids, saponins
Lagerstroemia speciosa	+	Temp 18-35 deg C	_	_	Terpenoids, glycosides, tannins
Lawsonia inermis L.	+	1.8-7.6m tall, temp 35-45 deg C	+	+	Tannins, terpenoids, coumarins
Manihot esculenta	+	Plant do not grow well at temp low than 60 deg C	_	_	_
Mimosa pudica	+	It usually grows 15- 50 cm tall, temp range of 60-85 F, 16-30C to germinate	+	+	saponins, coumarin, tannins
Nigella sativa L.	+	Altitude 220	+	_	Linoleic acid, palmitic acid
Olea europaea	+	It is grown between 30-45 deg latitude	_	_	Triterpenoids, betalains
Phyllanthus emblica L.	+	Annual daytime temp are within the range 20-29 deg C	_	_	Ethyl acetate

Piper betle L.	+	Altitude upto 900m, 22-27 deg C	_	+	saponins, tannins
Spondias pinnata	+	Altitude 1200m	_	_	Gallic acid
Syzygium aromaticum	+	Altitude 0-1000m, mean annual temp 25 deg C	_	+	Eugenol
Terminalia arjuna	+	25m tall, temp 20-33 deg C	_	+	saponins, glycosides, tannins
Tamarindus indica L.	+	Low to medium altitude 1000-1500m	_	_	Hexadecanol, pentadecanol
Zingiber officinale	+	Temp are within the range 19-29 deg C	_	_	Monoterpenoids, sesquiterpenoids

Objective 3.2: They also shows some other activity (Hepatoprotective Activity of medicinal plants) :

Procedure: Medicinal plants are important in human health care. 7,500 plants are thought to be used in local health rituals in rural areas. A thorough study and documentation of plants used in local health traditions as well as a pharmacological review of these plants and their taxonomic relatives, will aid in the development of valuable plant medicines for a range of ailments. Random plant sampling hasn't proven to be cost-effective.

9.5% of the plants were devoid of this activity and the action of these plants is largely attributed to their phytoconstituents such as antioxidant, flavonoids and anti-inflammatory effects.[21], [22].

Table2 : Different medicinal plants and (hepatoprotective activity)

Name of the plant	***Source of family***	***Plant part used***	***Hepatoprotective inducing agents***	***Extracts***
Baliospermum montanum	*Euphorbiaceae*	*Roots*	*Paracetamol*	*Alcohol, chloroform extract*
Cassia fistula	*Leguminosae*	*Leaf*	*Carbon tetrachloride*	*Methanol*
Aloe barbadensis Mill	*Liliaceae*	*Dried aerial plants*	*Carbon tetrachloride*	*Petroleum ether, chloroform, methanol*
Picrorrhiza rhizome	*Scrophulariaceae*	*Dried underground stem*	*Poloxamer-407*	*Water*
Azadirachta indica	*Meliaceae*	*Leaf*	*Paracetamol*	*70%ethanol*
Spondias pinnata	*Anacardiaceae*	*Stem heart wood*	*Carbon tetrachloride*	*Ethyl acetate, methanolic*
Acacia catechu	*Leguminosae*	*Powder plate catechu*	*Carbon tetrachloride*	*Ethyl acetate*

Chapter 4

Results and discussion

4.1 List of Plants found Himachal Pradesh with their different therapeutic activity and bioactive compounds:

Botanical name of the plant	Alkaloid compound	Flavonoid compound	Another chemical compound	Plant part used	No. of times compound activity was reported	Other therapeutic activities	References
Allium sativum	_	+	Vinyldithiins	Bulb	The constituents were tested more than 40 times	Antibacterial, antifungal,antiviral, antihypertensive	[23], [24]
Althaea cannabina	_	+	Phenolic acids, sucrose	Leaves	Constituents were tested 34 times	Antibacterial activity, antimicrobial	[25], [26]
Allamanda schotii	_	+	_	Flower	Constituents were tested 12 times	Antimicrobial activity, anti-inflammatory	[27]
Aloe barbadensis	_	+	Caffeic acids	Leaves	Constituents were tested 37 times	Gastro-protective,antifungal, anti-inflammatory properties	[28]
Aconitum ferox	+	_	Indaconitine	Flower	Constituents were tested 12 times	Anti-pyretic, analgesic, anti-rheumatic	[29]

						properties	
Alstonia scholaris	_	_	Beta-sterol	Leaves, seeds, bark	Constituents were tested 40 times	Antibacterial properties	[30]
Bacopa monnieri	+	_	Nicotinine	Flower	Constituents were tested more than 30 times	Antioxidant properties ,,anticancer properties	[31]
Bauhinia vahlii	_	+	Tannins	Stems, leaves	Constituents were tested 52 times	Antibacterial activity	[32]
Curcuma longa	_	_	Polyphenolic	Stem	Constituents were tested more than 20 times	Hepatic disorders, antiungal properties , anticancer	[33]
Cinnamous Zeylanicum	_	_	Cinnamyl acetate	bark	Constituents tested more than 40	Antioxidant, antimicrobial , anti-proliferative	[34]
Coriandrum sativum	_	_	Dodecanal	Leaves, seeds	Constituents test 20 times	Anti-diabetic properties	[35], [36]
Citrus limon L.	_	+	Citronellal	Oil	Constituents tested 8 times	Anticancer	[37], [38]
Carica papaya	+	_	Pseudocarpaine	Seeds	Constituents tested more than 40 times	Antimalarial, antioxidant, diuretic, anti-inflammatory	[39], [40]

Camellia sinensis	_	_	Terpenoids, glycosides	Leaves	Constituents tested 40 times	Antioxidant	[41]
Dillenia indica L.	+	+	Saponins, tannins	Barks, leaves	Constituents tested 40 times	Asthma, influenza, rheumatic pain	[42]
Dolichos biflorus	+	+	Tannins	Seeds	Constituents tested 11 times	Antipyretic, diuretic	[43]
Datura metel	_	_	Scopolamine	Seeds	Constituents tested more than 18	Analgesic, anti-inflammatory, anthelmintic	[44], [45]
Eucaplytus globules	_	_	Alpha-pinene	Leaf	Constituents tested more than 40	Arthritis, skin problems	[46], [47]
Eclipta alba (L). Hassk	_	+	Terpenoids	Leaf, flower, root, stem	Constituents tested more than 40	Anticancer, antifungal, insecticidal properties	[48]
Ficus carica	+	_	Terpenoids, eugenol	Fruit, root, leaves	Constituents tested 40 times	Cardiovascular disorders, anti-inflammatory	[49], [50]
Ficus racemosa	_	_	Tannins	Leaves, fruit	Constituents tested 46 times	Inflammatory, liver disorders, diabetes	[51], [52]

Gmelina arborea	_	+	Lignans	Root, bark	Constituents tested 16 times	Blood purifier, cough, wounds, ulcers	[53]
Hibiscus rosa-sinensis L.	+	+	Steroids, saponins	Leaves	Constituents tested more than 20 times	Inflamation, diabetes	[54]
Lagerstroemia speciosa	_	_	Terpenoids, glycosides, tannins	Leaves	Constituents tested 40 times	Antioxidant, antidiabetic, anti-diuretic	[55]
Lawsonia inermis L.	+	+	Tannins, coumarin	Flower,root, stem, leaves, seeds, bark	Constituents tested 17 times	Antioxidant, antidiabetic, hepatoprotective, anticancer	[56]. [57]
Manihot esculenta	_	_	Phenolic	Roots, leaves	Constituents tested 40 times	Headache, hypertension	[58]
Mimosa pudica	+	+	Saponins, tannins	Roots, leaves	Constituents tested 85	Antibacterial, antidepressant	[59], [60]
Nigella sativa L.	+	_	Palmitic acid	Seed	Constituents tested 45	Diuretic, analgesic, anti-inflammatory, hepatoprotective	[61]
Olea europaea	_	_	betalanins	Leaves	Constituents tested 40	Diuretic, laxative, skin cleanser	[62]
Phyllanthus	_	_	Ethyl acetate	Fruit, seeds,	Constituents tested	Antioxidant, anti-	[63], [64]

emblica L.				leaves, bark	60 times	inflammatory, antipyretic, hepatoprotective	
Piper betle L.	_	+	Saponins, tannins	Leaves	Constituents tested 50 times	Digestive	[65], [66]
Spondias pinnata	_	_	Gallic acid	Fruits, leaves, bark	Constituents tested more than 20 times	Anti-tubercular agent	[67]
Syzygium aromaticum	_	+	Eugenol	Flower	Constituents tested 40 times	Analgesic, antioxidant, antiviral, anti-inflammatory,antibacterial	[68]
Terminalia arjuna	_	+	Saponins, tannins	Bark	Constituents tested more than 40 times	Antioxidant, anti-inflammatory, anti-carcinogenic, gastro-productive effect	[69]
Tamarindus indica L.	_	_	Hexadecanol, pentadecanol	Dried fruit	Constituents tested 20 times	Parasitic, wound healing, respirator problems	[70]
Zingiber officinale	_	_	Monoterpenoids	Roots	Constituents tested 30 times	Antidiabetic, anticancer, antiarthritis	[71]

Chapter 5

Conclusion

Medicinal plants play vital roles in disease prevention and their use all existing prevention strategies like Aloe, Turmeric, Ginger and many other plants cure several common ailments and maximum plants were use for cold, cough, wounds, inflammation and some plants species in addition to their medicinal importance are religious and cultural importance. These medicinal plants can be used in the traditional way and also be incorporated in the modern medicine to benefit a large section of the society.

Chapter 6

References

[1] Badola HK. Medicinal plant diversity of Himachal Pradesh. In: Samant SS, Dhar U and Palni LMS(eds), Himalayan Medicinal Plants: Potential and Prospects. Nainital: Gyanodarya Prakashan; 2001:87-116

[2] Chauhan NS. Medicinal Orchids of Himachal Pradesh. Journal of Orchids Society of India 1990;4:99-105

[3] Badola HK and Pal M. Threatened medicinal plants and their conservation in Himachal Himalaya. Ind For 2003;129:55-68

[4] Mans DRA. From forest to pharmacy: plant-based traditional medicines as source for novel therapeutic compounds. Acad J Med Plants.2013; 1:101-10.

[5] Colegate SM, Molyneux RJ (2007) Bioactive natural products: detection, isolation, and structural determination. CRC Press, Boca Raton, pp 421-437

[6] Chauhan, N.S. (2003). Important medicinal and aromatic plants of Himachal Pradesh. Indian Forester,129(8), 979-998.

[7] Yuan H, Ma Q, Ye L, Piao G. The traditional medicine and modern medicine from natural products. Molecules. 2016;21(5):559.

[8] BarrataT.M., Dorman, H.J.D. et al.,.1998 Antimicrobial and antioxidant properties of some commercial essential oils. Flavour Fragr. J. 13:335-244.

[9] Abebe, B. A., & Chane Teferi, S. (2021). Ethnobotanical study of medicinal plants used to treat human and livestock ailments in Hulet Eju Enese Woreda, east Gojjam zone of Amhara region, Ethipia. Evidence-Based Complementary and Alternative Medicine: ECAM, 2021, 6668541.

[10] Rasool H.B. Medicinal Plants. Importance and Uses. Pharmaceut. Anal. Acta. 2012;3:e139. doi: 10.4172/2153-2435.1000e139.

[11] Heimenz JW, Walsh TJ. Lipid formulations of amphotericin B: recent progress and future directions. Clin Infect Dis 1996; 22Suppl. 2: S133-44.

[12] D.L. Mayers, S.A. Lerner, M. Ouelette, et al. Antimicrobial Drug Resistance C: Clinical and Epidemiological Aspects, vol. 2, Springer Dordrecht Heidelberg, London (2009) pp. 681-1347.

[13] Jorgensen J.H., Ferraro M.J. Antimicrobial susceptibility testing: a review of general principles and contemporary practices. Clin. Infect. Dis. 2009;49:1749-1755.

[14] Vidal, P., Schwarz, P., & Dannaoui, E.(2019). Evaluation of the gradient concentration strip method for antifungal susceptibility testing of isavuconazole and comparators for Mucorales species. Antimicrobial Agents and Chemotherapy, 63(10).

[15] Bauer AW, Kirby WM, Sherris JC, Turck M (April 1966). "Antibiotic susceptibility testing by a standardized single disk method". American Journal of Clinical Pathology. 45 (4): 493-496.

[16] Gulhane, A.R., Giri, G.K., & Khambalkar, S. V. (2018). Antifungal activity of aroma chemicals against graminicolous seed borne fungi by poisoned food method. International Journal of Current Microbiology and Applied Sciences, 7(07), 4102-4107.

[17] N. E. Malliga, M. S. Dhanarajan, and I. Elangovan, "Evaluation of antibacterial and antifungal activity of Phyllanthus emblica leaf extract," International Research Journal of Pharmaceutical and Biosciences., vol. 2, no. 2, pp. 59-66, 2015.

[18] L. B. Gende, I. Floris, R. Fritz, and M. J. Eguaras, "Antimicrobial activity of cinnamon (Cinnamomum zeylanicum) essential oil and its main components against Paenibacillus larvae from Argentine," Bulletin of Insectology, vol. 61, no. 1, pp. 1–4, 2008.

[19] S. Tajbakhsh, K. Mohammadi, I. Deilami et al., "Antibacterial activity of indium curcumin and indium diacetylcurcumin," African Journal of Biotechnology, vol. 7, no. 21, pp. 3832–3835, 2008.

[20] G. G. Nascimento, J. Locatelli, P. C. Freitas, and G. L. Silva, "Antibacterial activity of plant extractsand phytochemicals on antibiotic-resistant bacteria," Brazilian journal of microbiology, vol. 31, pp.247-256,2000.

[21] Mujeeb, V. Aeri,, P. Bagri, S. A. Khan, Hepatoprotective Activity of Methanolic Extract of Tylophora Indica(Burm.F,) Merill. Leaves, International Journal Of Green Pharmacy, Oct-17 2010, Pg No-125-127.

[22] S. Pradhan and C. Girish, :Hepatoprotective herbal drug, silymarin from experimental pharmacology to clinical medicine," Indian Journal of Medical Research, vol. 124, no. 5, pp.491-504,2006.

[23] Bozin, B., N. Mimica-Dukic, I. Samojlik, A. Goran, and R. Igic. 2008. Phenolics as

antioxidants in garlic (Allium sativum L., Alliaceae). Food Chem. 111:925–929.

[24] Beato, V.M., F. Orgaz, F. Mansilla, and A. Montano. 2011. Changes in phenolic compounds in garlic (Allium sativumL.) owing to the cultivar and location of growth. Plant Foods Hum. Nutr. 66:218–223.

[25] Akabori Y and Hasagava M. Flavonoid pattern in the pteridaceae , Flavoniod consistituents in the frounds of Adiantum Capillus-Veneris and A. Cuneatum [J]. Shok Zas 1969; 82294-297.

[26] Balandrin MF, Klocke JA, Wutule ES, Bollinger WH (1985): Natural plant chemicals: Sources of industrial and medicinal materials. Science 228: 1154–1160.

[27] Ashrafuzzaman M., Ali H., Liza L.N., Zinnah K.M.A. Antimicrobial activity of some medicinal plants against multi drug resistant human pathogens. Adv. Biosci. Bioeng. 2013;1:1–24.

[28] Kalra M., Garg N., Rallan M., Pathivada L., Yeluri R. Comparative evaluation of fresh Aloe barbadensis plant extract and mineral trioxide aggregate as pulpotomy agents in primary molars: A 12-month follow-up study. Contemp. Clin. Dent.2017;8:106–111.

[29] Kim D. K., Kwon H. Y., Lee K. R., Rhee D. K. & Zee O. P. Isolation of multidrug resistance inhibitor from Aconitum. Arch Pharmacol Res 21, 344–347 (1998).

[30]) Prachayasittikul S., Saraban P., Cherdtrakulkiat R., Ruchirawat S., Prachayasittikul V. New bioactive triterpenoids and antimalarial activity of Diospyros rubra Lec. 2010;9:1–10.

[31] Joshi A.G., Pathak A.R., Sharma A.M., Singh S. High frequency of shoot regeneration on leaf explants of Bacopa monnieri. Environ. Exp. Biol. 2010;8:81–84.

[32] Achenbach H, Stocker M, Constenia MA. Flavonoid and other constituents of Bauhinia manca. Phytochemistry. 1988;27:1835–41.

[33] N. Chainani-Wu. (2003). Safety and anti-inflammatory activity of curcumin: a component of turmeric (Curcuma longa). J. Altern. Complement Med. 9, 161-8.

[34] Gende L. B., Floris I., Fritz R., Eguaras M. J. Antimicrobial activity of cinnamon (Cinnamomum zeylanicum) essential oil and its main components against Paenibacillus larvae from Argentine. Bulletin of Insectology. 2008;61(1):1–4.

[35] Bakkali F, Averbeck S, Averbeck D, Idaomar M (2008) Biological effects of essential oils - a review. Food Chem Toxicol 46: 446–475.

[36] Matasyoh JC, Maiyo ZC, Ngure RM, Chepkorir R (2009) Chemical composition and antimicrobial activity of the essential oil of Coriandrum sativum. Food Chem.113: 526–529.

[37] Pathak D., Pathak K., Singla A. K. Flavonoids as medicinal agents. Recent advances. Fitoterapia. 1991;62(5):371–389.

[38] Tripoli E., Guardia M. L., Giammanco S., Majo D. D., Giammanco M. Citrus

flavonoids: molecular structure, biological activity and nutritional properties: a review. Food Chemistry. 2007;104(2):466–479.

[39] Ramasawamy AS and Sirsi M: Antituberculosis Activity of Some Chemical Constituents from Higher Plants. Indian J. Pharm. 1960; 22: 34-35.

[40] Kovendan K: Antimalarial activity of Carica papaya(Family: Caricaeae) leaf extract against Plasmodium falciparum; Asian Pacific Journal of Tropical Disease 2012; 2(1): 306-311.

[41] Taiyu Ren, Pengcheng Zheng, Kexin Zhang, Jieren Liao, Fei Xiong, Qiang Shen, Yuanchun Ma, Wanping Fang, Xujun Zhu. Effects of GABA on the polyphenol accumulation and antioxidant activities in tea plants (Camellia sinensis L.) under heat-stress conditions. Plant Physiology and Biochemistry 2021, 159 , 363-371.

[42] Banerji N, Majumbder P, Dutta NI. A new pentacyclic triterpene lactone from Dillenia indica. Phytochemistry 1975; 14: 1447-48.

[43] Muthu AK, Sethupathy S, Manavalan R, Karar PK. Hypolipidemic effect of methanolic extract of Dolichos biflorus Linn. in high fat diet fed rats. Indian J Exp Biol.2005;43:522–525.

[44] Wannang N. N., Ndukwe H. C. (2009). Evaluation of the analgesic properties of the Datura metel seeds aqueous extract. J. Med. Plants Res. 4 192–195

[45] Ma C. Y., Williams I. D., Che C. T. (1999). Withanolides from Hyoscyamus niger seeds. J. Nat. Prod.62 1445–1447.

[46] Cimanga K et al.: Correlation between chemical composition and antibacterial activity of essential oils of some aromatic medicinal plants growing in the Democratic Republic of Congo. Ethnopharmacol. 2002; 79: 213-220.

[47] Benyahia S et al.: Cladocalol: a pentacyclic 28-nor-triterpene from *Eucalyptus cladocalyx* with cytotoxic activity. Phytochemistry, 2005; 66: 627-632.

[48] Adhirajan N, Dixit VK, Gowri C (2001) Development and evaluation of herbal formulations for hair growth. Indian Drugs 38(11):559–563

[49] M. A. Saeed and A. W. Sabir, "Irritant potential of triterpenoids from Ficus carica leaves," Fitoterapia, vol. 73, no. 5, pp. 417–420, 2002.

[50] F. Vallejo, J. G. Marín, and F. A. Tomás-Barberán, "Phenolic compound content of fresh and dried figs (Ficus carica L.)," Food Chemistry, vol. 130, no. 3, pp. 485–492, 2012.

[51] Charde RM, Dhongade HJ, Charde MS, Kasture AV. Evaluation of antioxidant, wound healing and anti-inflammatory activity of ethanolic extract of leaves of Ficus religiosa. Int J Pharm Sci Res. 2010;19(5):73–82

[52] Murti K, Lambole V, Gajera V, Panchal M. Exploration of healing promoting potentials of roots of Ficus religiosa. Pharmacologia. 2011;2(12):374–378.

[53] Tiwari N., Yadav A.K., Srivastava P., Shanker K., Verma R.K., Gupta M.M. Iridoid glycosides from Gmelina arborea. Phytochemistry. 2008;69:2387–2390.

[54] Sohel A, et al. Antibacterial activity of the ethanol extracts of Hibiscus rosa-sinensis leaves and flowers against clinical isolates of bacteria. Bangladesh J Life Sci. 2010;22(2):65–73.

[55] Gupta A., Agrawal V.K., Rao C.V. Exploration of analgesic and antiinflammatory potential of Lagerstroemia speciosa. J. Appl. Pharmaceut Sci 2017;7:156–161.

[56] Hussain T, Arshad M, Khan S, Sattar H, Qureshi MS. In vitro screening of methanol plant extracts for their antibacterial activity. Pak J Bot. 2011;43:531–538.

[57] Chaudary GD, Poonia P, Kamboj P, Kalia AN. Hepatoprotective protection of Lawsonia inermis L, (seeds) I J phytopharmacol. 2012;3:66–73.

[58] Evans Atwijukire, Joseph Ffuna Hawumba, Yona Baguma, Enoch Wembabazi, Williams Esuma, Robert Sezi Kawuki, Ephraim Nuwamanya. Starch quality traits of improved provitamin A cassava (Manihot esculenta Crantz). Heliyon 2019, *5* (2) , e01215.

[59] Feroz MR, Ahmad STAK, Sindhu, Shahbaz AM. Antifungal activities of saponins from indigenous plant roots. Pak. Vet. J. 1993;13:4.

[60] Rajendran R, Sundararajan R. Preliminary phytochemical analysis and antibacterial activity of Mimosa pudica Linn. Leaves. Int J Pharm Bio Sci. 2010;6:1-8.

[61] Atta Ur R., Malik S., Zaman K. (1992). Nigellimine: a new isoquinoline alkaloid from the seeds of Nigella sativa . J. Nat. Prod. 55, 676–678.

[62] Haloui E, Marzouk Z, Marzouk B, Bouftira I, Bouraoui A, Fenina N. Pharmacological activities and chemical composition of the Olea europaea L. leaf essential oils from Tunisia.J Food Agric Environ. 2010;8(2):204–208.

[63] Gessler M, Nkunya MH, Mwasumbi LB, Heinrich M, Tanner M. Screening Tanzanian medicinal plants for antimalarial activity.Acta Trop. 1994;56(1):65–77

[64] Adhikary P., Chowdhury D., Banerji J., Chatterjee A. Antifertility effect of crude alcoholic extract of Piper betle stalk. Ind. J. Physiol. Allied Sci.1998;52:22–27.

[65] Sharma, M.L., Kawat, A.K.S., Balasabrahmanyan, V.R., and Singh, A.: Studies on the essential oil of betle vine leaf (Piper betle).Indian Perfuner 27. 91 (1983).

[66] Muhammad A., Rahman M.S., Kabir A.H., Kabir S., Hossain M.K. Antibacterial and cytotoxic activities of Spondias pinnata(Linn. f.) Kurz fruit extract. Indian J. Nat. Prod. 2011;2:265–267.

[67] Zhang Y., Wang Y., Zhu X., Cao P., Wei S., Lu Y. Antibacterial and antibiofilm activities of eugenol from essential oil of Syzygium aromaticum (L.) merr. & L. M. Perry

(clove) leaf against periodontal pathogen porphyromonas gingivalis. Microbial Pathogenesis. 2017;113

[68] Byeol Ryu, Hye Mi Kim, Jeong-Hwa Woo, Jung-Hye Choi, Dae Sik Jang. A new acetophenone glycoside from the flower buds of Syzygium aromaticum (cloves). Fitoterapia 2016, 115, 46-51.

[69] Rao M, Kumar MM, Rao MA. In vitro and in vivo. effects of phenolic antioxidants against cisplatin-induced nephrotoxicity. J Biochem(Tokyo)1999;125:383–390.

[70] Bathaei H.S., Jahromi K.H. The effect of flavonoids catechin, baclofen and saclofen on acetic acid-induced visceral pain by in rat. Int J Pharma Res. 2016;5:81–85.

[71] Awan U.A., Ali S., Shahnawaz A.M., Shafique I., Zafar A., Khan M.A.R., Ghous T., Saleem A., Andleeb S. Biological activities of Allium sativum and Zingiber officinale extracts on clinically important bacterial pathogens, their phytochemical and FT-IR spectroscopic analysis. Pak. J. Pharm. Sci. 2017;30:729–745.